IN THE UNITED STATES PATENT AND TRADEMARK OFFICE

IN THE MATTER OF:

AOYAGI et al.

GROUP: 1645

SERIAL NO.: 09/269,897

EXAMINER: Robert Zeman

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CONFIRMATION NO.: 1769

FOR: METHODS FOR DETECTION OR MEASUREMENT OF VIRUSES

Commissioner for Patents
P.O.Box 1450
Alexandria, VA 22313-1450

DECLARATION UNDER 37 C. F. R. § 1.132

Sir:

The undersigned, as a co-inventor and applicant of the present invention, hereby declares that:

1. I hold a master degree from Tohoku University in pharmacy.

2. I have been employed by Advanced Life Science Institute, Inc. as Executive Scientist since 1999 in the R & D department. I have direct experience in biochemistry and am capable of rendering an opinion as an expert in the art of biotechnology.

3. I have reread the specification of the subject patent application and all of the arguments raised by the Examiner during the prosecution.

4. In Example 5, 100µl of a sample was mixed with 100µl of a treatment solution (5% SDS, 0.6% CHAPS, 6M urea, 0.75% Triton X-100), and after treatment at 56°C for 30 minutes, 120µl of a reaction buffer and 120µl of the treated sample were added to a well in which antibodies C11-3 and C11-7 were immobilized. For this solution, the volume of the reaction buffer is very conventional in the art, and one skilled in the art would not expect undue experimentation to be required to determine the volume of the reaction buffer.

5. Moreover, from Examples 4, 5, and 14, the specific compositions of the reaction buffer is not essential i.e., it would be understood by one skilled in the art that different compositions can be used to achieve the same result. More specifically, regarding the composition of the reaction buffer, Example 4 exemplifies as a reaction buffer "100mM sodium phosphate buffer, pH7.3, containing 0.15M NaCl, 1% BSA, 0.5% Casein Na, 0.05% Tween 20". To show that a specific composition of the reaction buffer is not required, an experiment was carried out, in which HCV core antigen assay was carried out in accordance with the procedures used in Example 5, using the same sample, but using 3 different reaction buffer compositions. The results are shown hereafter in the following Table A:

TABLE A

Treatment: SDS conc. In 1st reaction Reaction Buffer pH:	- 0% A 7.3	+ 1.25% A 7.3	+ 1.25% B 7.3	+ 1.25% C 7.2
Composition	100 mM Na-Pi 1% BSA 0.5%Casein Na 0.05% Tween 20 0.15M NaCl	100 mM Na-Pi 1% BSA 0.5% Casein Na 0.05% Tween 20 0.15M NaCl	100 mM K-Pi 0.1% BSA 0.05 Casein Na 20mM EDTA 0.15M NaCl 0.4% Triton X-100	100mM K-Pi 1% BSA 0.05% Casein Na 20mM EDTA 0.1M NaCl 0.5% Triton X-100 0.05% Tween 20
Sample				
Normal serum	0.002	0.004	0.006	0.004
HCV Panel serum 13	0.012	0.645	0.624	0.603
HCV Panel serum 44	0.000	0.047	0.043	0.064

Absorbance was measured at a wavelength of 492nm (OD492) with the absorbance at 630nm as a reference.

As can be seen from the above Table A, in a control assay using a treatment solution without SDS, measurement value of HCV Panel serum was as low as, and substantially the same as that of normal serum. On the other hand, in the case where treatment buffer containing SDS was used in the treatment step, and one of three reaction buffers was used in the reaction step, measurement values were substantially the same between the different reaction buffers and were higher than that of the normal serum. These results show that the specific composition of the reaction buffer is not essential. In addition, Example 10 exemplifies another reaction buffer.

Regarding Example 14, in the detection of HBV core antigen, the volume of the reaction buffer is conventional, and a specific volume is not required. In addition, the composition of the reaction buffer used in Example 14 is conventional. Therefore, a person with ordinary skill in the art can easily select a volume and a composition of the reaction buffer, for the assay of HBV.

6. Furthermore, to one skilled in the art, after reading Example 4, 5, and 14, it is self evident that the volume of the reaction buffer used in the detection of HBV core antigen is conventional and as explained above in connection with Example 14 a specific volume is not required nor is a specific composition required.

7. Any person with ordinary skill in the art after reading the subject specification can easily select a volume and a composition of the reaction buffer to be used for the assay of HBV particularly after reading Examples 4, 5, and 14.

8. It should be understood that the purposes of treatment in accordance with the present invention is: (1) to destroy virus and to release core antigen, and (2) to decompose and inactivate endogenous antibodies. For example, about 1.4 grams of SDS, which is a strong protein decomposition agent, will bind to 1 gram of protein. Since a concentration of proteins in a serum is about 50 to 100 mg/ml, in the case where 5% SDS (50mg/ml) is added to a sample at the 1:1 volume ratio, most of the added SDS may bind to the proteins in the sample. Therefore, after treatment and at the step of combining the treated sample and an antibody probe, a reaction mixture should not contain SDS in an amount sufficient to inactivate the antibody probe. In addition, the treated sample is diluted with a reaction buffer and therefore a concentration of the originally added SDS should become even lower. Accordingly, one skilled in the art would not believe that SDS will affect a reaction of the core antigen and the antibody probe.

9. I declare that all statements made herein of my own knowledge are true and that all statements made on information and belief are believed to be true; and further that these statements were made with the knowledge that willful false statements and the like so made are punishable by fine or imprisonment, or both, under Section 1001 of Title 18 of the United States Code, and that such willful false statements may jeopardize the validity of the application or any patent issuing thereon.

Signed and declared at: 2-10-28 Nunoyamadai, Wako, Saitama, Japan

(City and country)

Signed By: Katsumi Aoyagi

(Declarant's Name)

Dated: February 6, 2006